Safety Data Sheet

6-Thioguanine

Division of Safety
National Institutes
of Health

WARNING!

THIS COMPOUND IS ACUTELY TOXIC, POSSIBLY TERATOGENIC, AND PRODUCES CHROMOSOMAL ABERRATIONS. IT IS ABSORBED THROUGH THE INTESTINAL TRACT. AVOID FORMATION AND BREATHING OF AEROSOLS.

LABORATORY OPERATIONS SHOULD BE CONDUCTED IN A FUME HOOD, GLOVE BOX, OR VENTILATED CABINET.

AVOID SKIN CONTACT: IF EXPOSED, WASH WITH SOAP AND COLD WATER. AVOID WASHING WITH SOLVENTS AND EXPOSURE TO UV LIGHT. AVOID RUBBING OF SKIN OR INCREASING ITS TEMPERATURE.

FOR EYE EXPOSURE, IRRIGATE IMMEDIATELY WITH LARGE AMOUNTS OF WATER. FOR INGESTION, INDUCE VOMITING. DRINK MILK OR WATER. REFER FOR GASTRIC LAVAGE. FOR INHALATION, REMOVE VICTIM PROMPTLY TO CLEAN AIR. ADMINISTER RESCUE BREATHING IF NECESSARY. REFER TO PHYSICIAN.

IN CASE OF LABORATORY SPILL, WEAR PROTECTIVE CLOTHING DURING CLEAN UP. AVOID SKIN CONTACT OR BREATHING OF AEROSOLS. SEE CASTEGNARO ET AL., 1985 FOR DETAILS. DISPOSE OF WASTE SOLUTIONS AND MATERIALS APPROPRIATELY.

A. Background

6-Thioguanine (6-TG) consists of colorless needles, soluble in alkaline aqueous solution (but subject to autoxidation in such solutions). It is toxic and produces chromosomal aberrations; carcino-

Issued: 5/87

Prepared by the Environmental Control and Research Program

genicity and mutagenicity have not been demonstrated and there is only one mention of teratogenicity in the literature. After activation (conversion into one or more nucleotide derivatives) its mode of action consists of incorporation into cellular RNA and DNA and consequent inhibition of nucleic acid and protein synthesis. Its principal use is as an antineoplastic agent in the treatment (alone or in combination with radiotherapy and/or other antineoplastics) of acute lymphocytic and chronic myelogenous leukemias

Review articles include: Paterson and Tidd, 1975; McCormack and Johns, 1982.

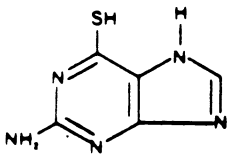
B. Chemical and Physical Data

Introductory note: 6-TG, when recrystallized from hot water, exist in the form of the hemihydrate. Unless specifically mentioned, numerical literature data on physical properties or dosages do not indicate whether reference is to the anhydrous or hydrated form.

1. Chemical Abstract No.: 154-42-7[A]

2. Synonyms: 2-Aminopurine-6-thiol; 2-amino-6-(1-H)-thione; purine-6-thiol-2-amino;[B] 6-mercaptoguanine; 6-H-purine-6-thione, 2-amino, 1,7-dihydro;[C] NSC-752.

3. Chemical Structure and Molecular Weight:



anhydrous form:	$C_5H_5N_5S$	167.2
hemihydrate:	$C_5H_5N_5S.1/2H_2O$	176.2

Note: The first of these two structures is the usually accepted form; however, for the closely related compound 6-mercaptopurine there is NMR evidence that at least the ribotide derivative exists i the second (thio) rather than the first (mercapto) form.

[A] Another CAS number, 1832-72-0, is found in CA Decennial Indices but has not been further identified. It is not listed in the CAS Registry Handbook.

[B] Chemical Abstracts name, used for listings in 6th to 8th Decennial Index.

[C] Chemical Abstracts name, used for listings in 9th Decennial Index and subsequently.

Density: No data.

Absorption Spectroscopy: Ultraviolet absorption maxima (ε) at pH 1: 258 (8,100) 347 (20,900) nm; at pH 11: 242 (8,700), 270 (7,200), 322 (16,000) nm (Elion and Hitchings, 1955); UV spectra at various pH values in the range 4.9-9.6 (Fox et al., 1958), the infrared spectrum (Sammul et al., 1964), and data on fluorescence at liquid nitrogen and room temperature (Al-Mosawi et al., 1980) have been published.

Volatility: No data; may be assumed to be low.

Solubility: No data; by analogy with the closely related 6-mercaptopurine (6-MP), 6-TG may be assumed to be practically insoluble in water, acetone, and ether; slightly soluble in dilute mineral acid (probably more so than 6-MP because of the 2-amino group) and dilute alkali (with autoxidation which may be prevented by addition of the antioxidant dithioerythritol or dithiothreitol).

Description: Colorless needles. Acid dissociation constants are $pKa_1 = 2.33$, $pKa_2 = 9.56$, $pKa_3 = >12$ (Bag et al., 1964).

Boiling point: No data; melting point: above 360°C.

Stability: Stable in solid form. Stable in dispersion in a suspending agent at room temperature for 84 days (less than 10% decomposition) (Dressman and Poust, 1983). Stability data in 0.01 N NaOH at 23°C have been published (Fox et al., 1958) but it is difficult to judge how rigorously atmospheric oxygen was excluded.

Chemical reactivity: The mercapto group is oxidized to sulfinate in alkaline iodine solution and to sulfonate in alkaline permanganate. Neutral iodine solution oxidizes 6-TG to the disulfide (Doerr et al., 1961). 6-TG forms chelates with divalent metal ions such as Pb, Zn, Ni, and Mn (Bag et al., 1964). For reactions in biological systems see F3.

Flash point: No data.

Autoignition temperature: No data.

Explosive limits in air: No data.

e, Explosion, and Reactivity Hazard Data

6-TG does not require special fire-fighting procedures or equipment and does not present unusual fire and explosion

2. No conditions contributing to instability are known to exist other than susceptibility to alkali and oxidizing agents.

3. No incompatibilities are known.

4. When heated to decomposition, 6-TG emits vapors of NO_x and SO_x.

5. 6-TG does not require non-spark equipment.

Operational Procedures

The NIH Guidelines for the Laboratory Use of Chemical Carcinogens describes operational practices to be followed when potentially carcinogenic chemicals are used in NIH laboratories. The NIH Guidelines should be consulted to identify the proper use conditions required and specific controls to be implemented during normal and complex operations or manipulations involving 6-TG.

It should be emphasized that this data sheet and the NIH Guidelines are intended as starting points for the implementation of good laboratory practices when using this compound. The practices and procedures described in the following sections pertain to the National Institutes of Health and may not be universally applicable to other institutions. Administrators and/or researchers at other institutions should modify the following items as needed to reflect their individual management system and current occupational and environmental regulations.

1. Chemical inactivation: Validated methods have been reported (Castegnaro et al., 1985).

2. Decontamination: Turn off equipment that could be affected by 6-TG or the materials used for cleanup. If there is any uncertainty regarding the procedures to be followed for decontamination, call the NIH Fire Department (dial 116) for assistance. Consult Castegnaro et al. (1985) for details concerning decontamination of surfaces, glassware, and animal cages.

3. Disposal: It may be possible to decontaminate waste streams containing 6-TG before disposal. For details, see Castegnaro et al. (1985). No waste streams containing 6-TG shall be disposed of in sinks or general refuse. Surplus 6-TG or chemical waste streams contaminated with 6-TG shall be handled as hazardous chemical waste and disposed of in accordance with the NIH chemical waste disposal system. Nonchemical waste (e.g., animal carcasses and bedding) containing 6-TG shall be handled and packaged for incineration in accordance with the NIH medical-pathological waste disposal system. Potentially infectious waste (e.g., tissue cultures) containing 6-TG shall be disinfected by heat using a standard autoclave treatment and packaged for incineration, as above. Burnable waste (e.g., absorbent bench top liners) minimally contaminated with 6-TG shall

be handled as potentially infectious waste and packaged for incineration, as above. Absorbent materials (e.g., associated with spill cleanup) grossly contaminated shall be handled in accordance with the chemical waste disposal system. Radioactive waste containing 6-TG shall be handled in accordance with the NIH radioactive waste disposal system.

4. Storage: Store solid 6-TG and its solutions in dark-colored, tightly closed containers, preferably under refrigeration. Avoid exposure to ultraviolet light and moisture. Store working quantities of 6-TG and its solutions in an explosion-safe refrigerator in the work area.

Monitoring and Measurement Procedures Including Direct Field Measurements and Sampling for Subsequent Laboratory Analysis

Introductory note: Several authors (Elion, 1967; McCormack and Johns, 1982) question the value of monitoring plasma levels of 6-TG since the compound is quickly transferred to intracellular sites and activated, and its effects become evident long after its disappearance from plasma. Nevertheless, analytical procedures for 6-TG are listed below, with emphasis on its biologically important metabolites.

1. Sampling: Plasma samples are deproteinized and extracted (particularly important if fluorimetry is used because of high background values). Low recoveries are considerably improved by conversion to the phenylmercury derivative (Dooley and Maddocks, 1980) and by addition of dithiothreitol prior to extraction (van Baal et al., 1984), or by ultrafiltration (Breithaupt and Goebel, 1981). Metabolites may be separated by solvent extraction (Dooley and Maddocks, 1982) or chromatography (Breter, 1977).

2. Analysis: The main methods are fluorimetry and high-performance liquid chromatography. Fluorimetry after oxidation to either the S-oxide (Thomas, 1976) or the sulfonate (Dooley and Maddocks, 1980) has been applied to plasma and urine, with a working range of 5-400 ng/ml, but suffers from variable recoveries and lack of specificity. HPLC has lower sensitivity (of the order of 0.2 µg/ml plasma) (Breithaupt and Goebel, 1981) but would be particularly useful in pharmacokinetic studies for the measurement of 6-TG and its metabolites (Breter, 1977; Andrews et al., 1982; van Baal et al., 1984). Special methods have been published for the analysis of 6-TG nucleotide (Lennard and Maddocks, 1983) and 6-methyl TG (Dooley and Maddocks, 1985).

ological Effects (Animal and Human).

Absorption: 6-TG is poorly and variably absorbed from the intestinal tract after oral administration (Ames et al., 1983); in spite of this, administration to patients is usually via this route. It is absorbed on parenteral injection.

Distribution and pharmacokinetics: No data.

Metabolism and excretion: The metabolism of 6-TG has been reviewed (Elion, 1967; Paterson and Tidd, 1975). Nelson et al. (1975) describe a metabolic scheme which should be consulted for details concerning enzymatic involvement in the various transformations. Briefly, the metabolism comprises anabolism sequentially to 6-TG-9 nucleoside, nucleotide, di- and triphosphate, and the corresponding deoxyribose derivatives, and catabolism which consists of the following three pathways: (a) conversion to guanine and sulfate, (b) oxidation with deamination to 6-thioxanthine and 6-thiouric acid, (c) S-methylation followed by the same reaction sequence outlined under anabolism. The triphosphates are incorporated into cellular RNA and DNA, a reaction which appears to constitute the mechanism of toxic and antineoplastic action of 6-TG; the main urinary excretion products are (in the dog) thiouric acid, sulfate and methylthioguanine, with small amounts of unchanged 6-TG, methyl thioxanthine, and other unidentified metabolites (Loo et al., 1981). Essentially the same distribution among urinary metabolites is found in man (Andrews et al., 1982).

Toxic effects: The acute single dose LD_{50} has been reported as follows (in mg/kg): mouse, 92 for male, 103 for female, ip; rat, over 250 for male, ip. The toxicity of 6-TG shows remark- ably high potentiation when given in repeated doses: for 5 daily doses the corresponding LD_{50}s (in mg/kg/day) are: mouse, 4.2 for male, 6.5 for female, ip; rat, 9.8 for male ip, 11.8 for male oral. In the dog the LD_{50} is approximately 0.6 mg/kg/day for 10 doses iv and about 2 mg/kg/day oral (Philips et al., 1956). This potentiation is considerably more pronounced than was found for 6-mercaptopurine (Philips et al., 1954). In further contrast with 6-mercaptopurine, single lethal doses are fatal within a few hours and there appears to be no production of hepatic lesions. Toxic symptoms in man and animals are leukopenia, thrombocytopenia, bone marrow depression, and some gastrointestinal disturbances (Philips et al., 1956; Paterson and Tidd, 1975). As stated above, the major biochemical lesion produced by 6-TG metabolites is due to their incorporation into tissue DNA and RNA and consequent disruption of normal nucleic acid and protein biosynthesis.

5. Carcinogenic effects: None has been reported.

6. Mutagenic and teratogenic effects: 6-TG is not mutagenic in the Ames test but produces chromosomal aberrations and breakages in Salmonella typhimurium (Yajima et al., 1981) and human cells in vitro (Engel et al., 1967). The only reference to possible teratogenic action mentions 75% fetal resorption in the rat on administration of 6-TG during gestation (Thiersch, 1962); it should be noted that 6-mercaptopurine is a teratogen in several animal species.

Emergency Treatment

1. Skin and eye exposure: For skin exposure, remove contaminated clothing and wash skin with soap and water. Skin should not be rinsed with organic solvents or scanned with UV light. Avoid rubbing of skin or increasing its temperature. For eye exposure, irrigate immediately with sodium bicarbonate solution, followed by copious quantities of running water for at least 15 minutes. Obtain ophthalmological evaluation.

2. Ingestion: Drink plenty of water or milk. Induce vomiting. Refer for gastric lavage.

3. Inhalation: Remove victim promptly to clean air. Administer rescue breathing if necessary.

4. Refer to physician. Consider treatment for pulmonary irritation.

References

Al-Mosawi, A.I., J.N. Miller, and J.W. Bridges. 1980. Determination of 6-mercaptopurine and related compounds by phosphorescence spectroscopy. Analyst 105:448-454.

Ames, M.M., G. Powis, and J.S. Kovach (eds.) 1983. Pharmacokinetics of Anticancer Agents in Humans. Chapter 15, Elsevier, NY.

Andrews, P.A., M.J. Gorin, M.E. May, and N.R. Bachur. 1982. Reversed-phase high-performance liquid chromatography analysis of 6-thioguanine applicable to pharmacologic studies in humans. J Chromatog 227:83-91.

Bag, S.P., Q. Fernando, and H. Freiser. 1964. Formation constants of certain metal complexes of thioguanine. Arch Biochem Biophys 106:379-380.

Breithaupt, H. and G. Goebel. 1981. Quantitative high pressure liquid chromatography of 6-thioguanine in biological fluids. J Chromatog Sci 19:496-529.

Breter, H.-J. 1977. The quantitative determination of metabolites of 6-mercaptopurine in biological materials. I. A separation method for purine and 6-thiopurine bases and nucleosides using high-pressure liquid-cation-exchange chromatography. Anal Biochem 80:9-19.

Castegnaro, M., J. Adams, M.A. Armour, J. Barek, J. Benvenuto, C. Confalonieri, U. Goff, S. Ludeman, D. Reed, E.B. Sansone, and G. Telling. 1985. Laboratory Decontamination and Destruction of Carcinogens in Laboratory Wastes: Some Antineoplastic Agents. IARC Scientific Publications No. 73. World Health Organization, Geneva, Switzerland.

Doerr, I.C., I. Wempen, D.A. Clarke, and J.J. Fox. 1961. Thiation of nucleosides. III. Oxidation of mercaptopurines. J Org Chem 26:3401-3409.

Dooley, T. and J.L. Maddocks. 1980. Assay of 6-thioguanine in human plasma. Brit J Clin Pharmacol 9:77-82.

Dooley, T. and J.L. Maddocks. 1982. Assay of an active metabolite of 6-thioguanine, 6-thioguanosine-5 -monophosphate, in human red blood cells. J Chromatog 229:121-127.

Dooley, T. and J.L. Maddocks. 1985. High-performance liquid chromatographic assay of 6-methylthioguanine, a major metabolite of 6-thioguanine, in urine. J Chromatog 337:321-327.

Dressman, J.B. and R.I. Poust. 1983. Stability of allopurinol and of five antineoplastics in suspension. Am J Hosp Pharm 40:616-618.

Elion, G.B. 1967. Biochemistry and pharmacology of purine analogs. Fed Proc 26:898-904.

Elion, G.B. and G.H. Hitchings. 1955. The synthesis of 6-thioguanine. J Am Chem Soc 77:1676.

Engel, W., W. Krone, and U. Wolf. 1967. Die Wirkung von Thioguanin, Hydroxylamin und 5-Brom-desoxyuridin auf menschliche Chromosomen in vitro [The effect of thioguanine, hydroxylamine and 5-bromo-desoxyuridine on human chromosomes in vitro.] Mutat Res 4:353-368.

Fox, J.J., I. Wempen, A. Hampton, and I.L. Doerr. 1958. Thiation of nucleosides. I. Synthesis of 2-amino-6-mercapto-9-β-D-ribofuranosylpurine ("Thioguanosine") and related purine nucleosides. J Am Chem Soc 80:1669-1675.

Lennard, L. and J.L. Maddocks. 1983. Assay of 6-thioguanine nucleotide, a major metabolite of azathioprine, 6-mercaptopurine and 6-thioguanine, in human red blood cells. J Pharm Pharmacol 35:15-18.

Loo, T.L., K. Lu, J.A. Benvenuto, and M.G. Rosen. 1981. Disposition and metabolism of thiopurines. III. β-2'-deoxythioguanosine and 6-thioguanine in the dog. Cancer Chemother Pharmacol 6:131-136.

McCormack, J.J. and D.G. Johns. 1982. Purine Antimetabolites. Chapter 9 in: Chabner, B. (ed.), Pharmacological Principles of Cancer Treatment. W.B. Saunders, Philadelphia, PA.

Nelson, J.A., J.W. Carpenter, L.M. Rose, and D.J. Adamson. 1975. Mechanisms of action of 6-thioguanine, 6-mercaptopurine, and 8-azaguanine. Cancer Res 35:2872-2878.

Paterson, A.R.P., and D.M. Tidd. 1975. 6-Thiopurine. in: Sartorelli, A.C. and D.G. Johns (eds.), Antineoplastic and Immunosuppressive Agents, Part II, Chapter 47. Handbuch der experimentellen Pharmakologie 38/2.

Philips, F.S., S.S. Sternberg, L. Hamilton, and D.A. Clarke. 1954. The toxic effects of 6-mercaptopurine and related compounds. Ann NY Acad Sci 60:283-296.

S.S. Sternberg, L.D. Hamilton, D.A. Clarke, B. Wheelock, and B. Bond. 1956. Effects of thioguanine in mammal Cancer 9:1092-1101.

Sammul, O.R., W.L. Brannon, and A.L. Hayden. 1964. Infrared spect of some compounds of pharmaceutical interest. J Assoc Off Anal Chem 47:918-991.

Thiersch, J.B. 1962. Effect of substituted mercaptopurines on the rat litter in utero. J Reprod Fertil 4:291-295.

Thomas, A.D. 1976. Spectrofluorometric determination of thiopurines. I. 6-thioguanine. Talanta 23:383-386.

vanBaal, J.M., M.B. van Leeuwen, T.J. Schouten, and R.A. de Abreu. 1984. Sensitive high-performance liquid chromatographic determination of 6-mercaptopurine, 6-thioguanine, 6-mercaptopurine riboside and 6-thioguanosine in biological fluids. J Chromatog 336:422-428.

Yajima, N., K. Kondo, and K. Morita. 1981. Reverse mutation tests in Salmonella typhimurium and chromosomal aberration tests in mammalian cells in culture on fluorinated pyrimidine derivatives Mut Res 88:241-254.